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CDK5RAP1 siRNA (m): sc-142226

BACKGROUND

Cyclin dependent kinase 5 (Cdk5) is a key regulator of cell cycle progression in neuronal differentiation that physically associates with and is activated by the neuron-specific protein p35. CDK5RAP1 (Cdk5 regulatory subunit-associated protein 1), also known as Cdk5 activator-binding protein C42, is a 601 amino acid protein that specifically inhibits Cdk5 activation by p35 through formation of a dimer that inhibits kinase activity. CDK5RAP1 contains one TRAM domain, which is thought to bind tRNA and deliver the RNA-modifying enzymatic domain to its target. There are 4 named isoforms of CDK5RAP1 that are produced as a result of alternative splicing events and are expressed at high levels in heart and skeletal muscle.

REFERENCES

1. Tang, D., et al. 1995. An isoform of the neuronal cyclin-dependent kinase 5 (Cdk5) activator. *J. Biol. Chem.* 270: 26897-26903.
2. Chin, K.T., et al. 1999. Identification and structure characterization of a Cdk inhibitory peptide derived from neuronal-specific Cdk5 activator. *J. Biol. Chem.* 274: 7120-7127.
3. Ching, Y.P., et al. 2000. Cloning of three novel neuronal Cdk5 activator binding proteins. *Gene* 242: 285-294.
4. Wang, X., et al. 2000. Identification of a common protein association region in the neuronal Cdk5 activator. *J. Biol. Chem.* 275: 31763-31769.
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6. Online Mendelian Inheritance in Man, OMIM[™]. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 608200. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
7. Zou, X., et al. 2004. Cloning, characterization and expression of CDK5RAP1_v3 and CDK5RAP1_v4, two novel splice variants of human CDK5RAP1. *Genes Genet.* 79: 177-182.

CHROMOSOMAL LOCATION

Genetic locus: Cdk5rap1 (mouse) mapping to 2 H1.

PRODUCT

CDK5RAP1 siRNA (m) is a pool of 3 target-specific 19-25 nt siRNAs designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see CDK5RAP1 shRNA Plasmid (m): sc-142226-SH and CDK5RAP1 shRNA (m) Lentiviral Particles: sc-142226-V as alternate gene silencing products.

For independent verification of CDK5RAP1 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-142226A, sc-142226B and sc-142226C.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20° C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20° C, avoid contact with RNases and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

CDK5RAP1 siRNA (m) is recommended for the inhibition of CDK5RAP1 expression in mouse cells.

SUPPORT REAGENTS

For optimal siRNA transfection efficiency, Santa Cruz Biotechnology's siRNA Transfection Reagent: sc-29528 (0.3 ml), siRNA Transfection Medium: sc-36868 (20 ml) and siRNA Dilution Buffer: sc-29527 (1.5 ml) are recommended. Control siRNAs or Fluorescein Conjugated Control siRNAs are available as 10 μ M in 66 μ l. Each contain a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA. Fluorescein Conjugated Control siRNAs include: sc-36869, sc-44239, sc-44240 and sc-44241. Control siRNAs include: sc-37007, sc-44230, sc-44231, sc-44232, sc-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

GENE EXPRESSION MONITORING

CDK5RAP1 (D-1): sc-398764 is recommended as a control antibody for monitoring of CDK5RAP1 gene expression knockdown by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500).

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgG κ BP-HRP: sc-516102 or m-IgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker[™] Molecular Weight Standards: sc-2035, UltraCruz[®] Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG κ BP-FITC: sc-516140 or m-IgG κ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz[®] Mounting Medium: sc-24941 or UltraCruz[®] Hard-set Mounting Medium: sc-359850.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor CDK5RAP1 gene expression knockdown using RT-PCR Primer: CDK5RAP1 (m)-PR: sc-142226-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.